THE PRESENT STATUS OF THE TREATMENT OF DIABETES WITH INSULIN *

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Progress in the treatment of diabetes mellitus is moving so rapidly that new experimental evidence to be of particular value must be published promptly. Because of this fact, and the care with which the work was done, the editor has acceded to the request of the Section on General Medicine and is publishing the article by Doctor Sansum and his co-workers in an earlier issue than it could be published under the usual conditions governing priority of publication.—*Editor.*

The work of Banting and Best and their associates in the isolation and clinical use of the sugar-metabolizing hormone, insulin, and its specificity in the treatment of diabetes is now well known and accepted. For a detailed account of the methods used and the results obtained by us in the first one hundred cases, the reader is referred to an article which recently appeared in the Journal of Metabolic Research. It is the purpose of this paper to present some of the newer developments of this problem.

I. A SMALL INSULIN FRACTION WHICH IS ACTIVE WHEN GIVEN TO RABBTS ORALLY IN WATER SOLUTION

Murlin, Clough, Gibbs, and Stokes have shown that the respiratory quotient could be raised when extracts of the pancreas dissolved in N/20 Na OH, together with glucose, were given by stomach tube. The alkali was given for the purpose of inactivating pepsin.

Winter used a dilute alcohol solution because alcohol is known to be absorbed from the stomach. He used a crude insulin and reported both significant drops in blood sugar and collapse of the animals. He failed to note drops in blood sugar in other types of solutions. Using the insulin prepared in the routine way, we have failed as yet to confirm Winter's findings.

For over a year, we have been using a large portion of our experimental product in attempts at effectual oral administration. In our first efforts, we endeavored to circumvent the action of the digestive enzymes, assuming that failure by the oral route was due to the destruction of the insulin by the digestive enzymes. We worked with insulin in various solvents and in different types of coated capsules, using as high as 1000 units per dose, but all of this work was frankly negative. It, therefore, seemed to us that some other hypothesis should be followed.

We are now assuming that the present insulin is impure and that protein substances containing it, or to which it is attached, are too large to pass through the intestinal wall without radical digestive changes, in which process the insulin is incidentally destroyed.

Macleod reported that from fish glands he was able to obtain an insulin which was biuret-free. There was considerable discussion at the meeting of the American Society of Biological Chemists, held in St. Louis, December 27-29, 1923, as to whether insulin was actually biuret-free. None of our purest insulin has been biuret-free, and the purest Lilly iletin gives a biuret reaction. Murlin believes that pure insulin will probably be biuret-free. We have spent considerable time in attempting to prepare a biuret-free insulin. We have hydrolized routine insulin with various strengths of acid and produced a biuret-free final product, but as yet this final product has had no insulin activity.

Following this line of thought, we have searched for a fraction of probably smaller molecular weight than the routine, insulin-containing material. The major portion of insulin is precipitated out in an alcohol percentage below 93 per cent. One of us, Maxwell, has isolated a small fraction that is soluble in 98.5 per cent alcohol and perhaps at a still higher percentage. We have found that this fraction of insulin is active when given to rabbits, orally, in water solution. It produces typical falls in biood sugar and convulsions. The dose necessary is comparatively large, but it may be that even this product contains considerable of the orally inactive insulin.

Exp. No. 1

Wt. 1.46 kg. Dose: 112 units given in 45 cc. of 10 per cent alcohol.

When taken	Blood sugar per cent
Initial	.105
End of first hour	.057
End of second hour	.069
End of third hour	.114

Exp. No. 2

Wt. 3.46 kg. Dose: 66 units given in 45 cc. water.

When taken	Blood sugar per cent
Initial	.108
End of first hour	.055
End of second hour	.055
End of third hour	.095

Exp. No. 3

Wt. 1.13 kg. 900 units in 45 cc. water.

When taken	Blood sugar per cent
Initial	.087
End of first hour	.091
End of second hour	.033
End of third hour	.085

Exp. No. 4

Wt. 1.58 kg. 800 units of material used in Exp. 3 in 40 cc. water.

When taken	Blood sugar per cent
Initial	.121
End of first hour	.055
End of second hour	.045

Typical convulsion 10 minutes later.

* Presented to the General Medicine Section at the Fifty-third Annual Session of the California Medical Association, Los Angeles, May, 1924.

Exp. No. 5. Control

Wt. 1.64 kg. 830 units of crude insulin.

When taken	Blood sugar per cent
Initial	.137
End of first hour	.143
End of second hour	.144
End of third hour	.137

We are now attempting, by various chemical means, to ascertain how much of the insulin made in the routine way can be changed into the orally active type. It may be that the drastic methods now used in the preparation of the present insulin will need to be changed to less drastic ones, lest in the process of preparation we are building up too large aggregations of molecules.

II. THE VALUE OF INSULIN ON DIFFERENT DIETS

There has been and still is considerable discussion as to whether the insulin unit has a constant sugar-burning power. Allen believes that the unit has a different value in each patient, and even in the same patient when different diets are used. We have believed from the beginning—and clinical usage has confirmed this belief—that when a patient is kept continuously "sugar-free" the unit has a constant value in patients of widely varying degrees of severity, and on widely different diets when such diets are reduced to the common denominator, sugar-former. The following experiment was planned to ascertain the value of the insulin on different diets. For a long time, this patient had been maintained continuously sugar-free on a diet containing carbohydrate, 91 grams; protein, 79 grams; fat, 193 grams; calories, 2417, with a diet "G" of 156 grams. One of us, Blatherwick, suggested that we vary the elements of this diet as widely as possible, keeping the calories and the "G" constant. For over three years, and in the pre-insulin days determined by diet alone, this patient has had a constant tolerance sufficient to metabolize 52 grams of sugar formers. He has had insulin continuously for nearly two years. He has now gained in weight from 95 pounds to his normal weight of 154 pounds, and his diet was reduced October 30, 1923, to 2417 calories, which is his maintenance level, since he is now neither losing nor gaining weight. The following tables show that we have varied the carbohydrate in his diet from 54 to 113 grams, the protein from 40 to 145 grams, and the fat from 180 to 200 grams, and that throughout the period of the experiment, twenty-three days, he continued to pass subnormal amounts of sugar in the urine. At no time during the course of the experiment was he too sugar-free.

Period I

C.	P.	Diet F.	Cal.	"G"	Tolerance "G"	Insulin "G"	Insulin Kilo Units
54	145	180	2416	156	52	104	80

Day	Vol.	Qual.	Per ct.	Grams	O. A.	N.
1	2050	— +	0.044	0.910	902	14.76
2	1551	— +	0.034	0.550	806	15.66
3	1500	— —	0.045	0.670	924	20.85
4	1850	— +	0.060	1.110	829	18.87
5	1700	— —	0.044	0.750	734	20.74
6	2150	— —	0.039	0.830	757	19.56
7	1400	— —	0.048	0.670	761	20.40
8	1750	— —	0.037	0.650	798	20.87
				Av. .767		

Insulin No. 246. About 20 units per day recovered in urine.

Period II

C.	P.	Diet F.	Cal.	"G"	Tolerance "G"	Insulin "G"	Insulin Kilo Units
113	40	200	2412	156	52	104	80

Day	Vol.	Qual.	Per ct.	Grams	O. A.	N.
1	1900	— +	0.033	0.620	623	12.16
2	1700	— +	0.033	0.560	653	7.90
3	1365	— —	0.036	0.490	842	5.24
4	1050	— +	0.043	0.450	639	5.05
5	2200	— —	0.029	0.630	756	5.97
6	1860	— —	0.030	0.560	669	4.94
7	1850	— —	0.031	0.570	666	4.97
8	1450	— —	0.041	0.600	765	4.71
				Av. .560		

Insulin No. 246. None recovered.

Period III

C.	P.	Diet F.	Cal.	"G"	Tolerance "G"	Insulin "G"	Insulin Kilo Units
91	79	193	2417	156	52	104	80

Day	Vol.	Qual.	Per ct.	Grams	O. A.	N.
1	2900	— —	0.022	0.650	626	6.26
2	1350	— —	0.040	0.540	616	6.91
3	1900	— —	0.032	0.600	790	9.84
4	2275	— —	0.023	0.520	736	10.12
5	1980	— —	0.029	0.570	649	9.00
6	1500	— —	0.042	0.630	708	9.03
7	1650	— —	0.030	0.500	554	7.75
				Av. .573		

Insulin No. 255. None recovered.

We have raised and lowered the caloric value of diets in a long series of cases, adjusting the insulin dosage to correspond to the change in the diet "G," with practically no errors in the estimation of the proper dosage; however, quantitative metabolic work like the above has not as yet been done in such cases. In the series now in progress, we are keeping the diet "G" constant, and varying the caloric value of the diet chiefly by the omission of fat, since Allen claims that the proper insulin dosage is more dependent upon the caloric content than upon the elements of the diet.

III. A PROBABLE CAUSE OF THE IRRITATING EFFECTS OF ILETIN

From the beginning, iletin has been more or less irritating. We formerly worked on the assumption that this irritation was due to the acidity of the iletin, since many lots had a pH of 4.4 to 4.0 or less. For a long time, we attempted to keep the reaction of our product at as close to the neutral point as possible. Insulin is soluble in water solution at a pH of 6.8, but is slowly destroyed at a pH of above 7.0, and insoluble at a pH of 6.6, making the margin of stability very narrow. We have lost many lots of insulin, either from decreases in potency or from precipitation. Some of these losses, we theoretically explained on the basis of minute solution of the soft glass container, thereby increasing the alkalinity of the solution, and others we felt were due to the introduction of slight traces of carbonic acid, which were introduced into the bottle with repeated fillings of the syringe. We, therefore, found it practically impossible to prepare a stable insulin at a pH of 6.8. In the beginning, our then very impure insulin was made up in normal salt solution, and we found such solutions hypertonic. We, therefore, dissolved the insulin in distilled water. After our insulin became less impure, we found that we had a hypotonic solution, and, on examining the iletin, we found it also a hypotonic solution. We, therefore, made up a lot of insulin in physiological salt soluton at a pH of 3.8 to 4, and found it not only stable, but non-irritating. We believe that the irritating effects of the iletin are due,

in part at least, to the fact that the solution is hypotonic.

IV. THE PRESENT VALUE OF THE ILETIN UNIT WITH A DISCUSSION FAVORING THE INTRAVENOUS INSTEAD OF THE SUBCUTANEOUS METHOD OF RABBIT EVALUATION

The variation in the clinical strength of the iletin has and is causing considerable trouble. The following quotation from a letter of Dr. Clowes of Eli Lilly & Company, dated April 2, will convince you that Eli Lilly & Company are fully in accord with our findings of variations in the clinical strength.

"As regards lots 746,255, which you approve as being fully up to strength, we are inclined to believe that this lot is somewhat stronger than the indicated unitage based on animal tests. As regards lot 741,138, of which you complained in a previous letter, we believe that the lot in question was somewhat below the indicated unitage. (We found that lot 746,255 had a clinical value of 1 gram per unit or 40 grams per cc. of the U-40 product, and that lot 741,138 had a clinical value of .75 grams per unit or 30 grams per cc. of the U-40 product.) Both these lots were carefully tested in Indianapolis and Toronto, and were approved at both points as being within 10 per cent of the desired unitage. But, as I have repeatedly stated to you in our correspondence, the difficulties in the way of making these tests are so great that it is almost impossible, in the light of our present knowledge, to guarantee the unitage to as close a point as 10 per cent variation above or below the desired figure."

In our experience in routine lots of iletin, the clinical sugar-burning power has varied, during the past six months, from 0.6 to 1 gram per unit on the same patient taking the same diet. However, the iletin which we have received during the past two months has been worth fully 1 gram per unit or 20 grams per cc. of the U-20 and 40 grams per cc. of the U-40 product.

We had the same difficulty in the evaluation of our insulin in the beginning. Because it is well known that wide variations occur in other types hypodermic of medication, we abandoned, July, 1922, the subcutaneous route as a method of evaluation of insulin.

Our rabbits are kept on an alfalfa diet. Page has shown that the convulsion dose is increased by acid dietaries, and we have been able to confirm these results. The alfalfa diet is a basic diet. The rabbits are used only once per week. If a convulsion does not follow the experimental dose, additional insulin is given that day sufficient to produce a convulsion. This convulsion is antidoted by glucose given subcutaneously, and for a day the rabbit is fed crushed barley. Unless fed with a concentrated carbohydrate food following a convulsion, the animals do not thrive. Our unit is the smallest amount of insulin per kilogram, which, when given intravenously, will cause a convulsion. We find the approximate convulsion dose roughly, and then use a number of rabbits, not more than ten, very close to this range. Our routine yield is so constant that we can often predict the potency of the insulin with a fair degree of accuracy. Such an intravenous rabbit unit has a very constant clinical value in the neighborhood of 1.3 grams. The minimum value is 1.2 grams, and the maximum 1.4 grams. More insulin is lost in the urine by this intravenous method than by the subcutaneous route; hence, this unit is higher in value than the subcutaneous unit.

SUMMARY

1. Evidence has been submitted showing that we have isolated a small fraction of insulin that is active by mouth.

2. Insulin has a constant sugar-metabolizing value on widely different diets.

3. We believe that the present irritating effect of the iletin may be due to the hypotonicity of the solution.

4. In our experience, the value of the iletin unit has varied during the past six months from .6 to 1 gram or from 12 to 20 grams per cc. of the U-20, and 24 to 40 grams per cc. for the U-40 product. Intravenous rabbit units show a more constant value.

The Value of Milk Acidified With Lemon Juice—Alfred F. Hess and Milton J. Matzner, New York (Journal A. M. A.), add fruit juices directly to the milk formulas, instead of giving them to the infants separately and between feedings. Their object in diverging from this practice was twofold—to simplify the technic of feeding, and to render the milk more acid. Lemon juice or orange juice can be added directly to cow's milk without bringing about curdling. By mixing approximately 21 cc. of lemon juice with a quart of milk, its buffer action is markedly reduced and the hydrogen-ion concentration increased from pH 6.64 to about 5.54. In this way, cow's milk is rendered more digestible, and its true acidity in the stomach is made to resemble more nearly that of human milk. Infants who received milk prepared with lemon juice thrived well for long periods. Lactic acid or hydrochloric acid have been added to cow's milk, with the same object in view. One advantage of using lemon juice for this purpose is that it also supplies antiscorbutic vitamin, thus compensating for the deficiency of this essential factor in milk. Egg yolk can be combined with the mixture of milk and lemon juice with but slight alteration of the hydrogen-ion content. This combination is well borne by infants. By this means, a food is prepared which compensates for the nutritional deficiencies of cow's milk, furnishing both the antiscorbutic and the antirachitic factors, as well as additional fat-soluble vitamin and iron.

Duodenal Ulcer—Victor Knapp, New York (Journal A. M. A.), asserts a niche and incisura visualized in the first portion of the duodenum is absolute proof of the presence of duodenal ulcer. The significance of this roentgen-ray sign of ulcer of the duodenum has received ample recognition abroad, while here in America it has been studied but little, with perhaps insufficient emphasis on its importance. Knapp cites one case in support of this contention.

Medical Honesty—Sir Thomas Browne reminds us in eloquent words that there is only one kind of honesty. "Live by old ethicks," he says, "and the classical rules of honesty. Put no new names or notions upon authentic virtues and vices. Think not that morality is ambulatory; that vices in one age are not vices in another; or that virtues which are under the everlasting seal of right reason may be stamped by opinion."